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Pressure Probe Technique for Measuring Water Relations of Cells in Higher Plants[1]

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ABSTRACT

A new method is described for continuously measuring cell turgor pressure (P), hydraulic conductivity (L_p), and volumetric elastic modulus (ϵ) in higher plant cells, using a pressure probe. This technique permits volume changes, ΔV, and turgor pressure changes, ΔP, to be determined with an accuracy of 10^{-5} to 10^{-6} μl and 3 to $5 \cdot 10^{-2}$ bar, respectively.

The main principle of the new method is the same as the pressure probe developed by Zimmermann and Steudle in which pressure is transmitted to a pressure transducer by means of an oil-filled capillary introduced into the cell. In order to use the pressure probe for small tissue cells, the effective compressible volume of the apparatus has to be sufficiently small in comparison to the volume of the cell itself. This is achieved by accurately fixing the oil/cell sap boundary in the very tip of the microcapillary by means of an electronic feedback mechanism, so that the effective volume of the apparatus is reduced to about 2 to 10% of the cell volume. In this way also, errors arising from compressibility of the apparatus and temperature fluctuations can be excluded.

Measurements on tissues cells of *Capsicum annuum* fruits yield ϵ values of 2 to 25 bar. Furthermore, ϵ can be shown to be a function of both cell turgor pressure and cell volume; ϵ increases with increasing turgor pressure and is higher in larger cells.

Direct measurements of cell turgor pressure, cell wall elasticity, and hydraulic conductivity of cell membranes in single cells of higher plants are crucial for the evaluation of water relations of higher plants and for the development of models for water transport in plant tissues. A number of attempts have been made to determine turgor pressure, the elastic modulus, and the hydraulic conductivity (water permeability) of cells in plant tissue (see reviews (3, 18, 19, 23)). In most cases, P[2] and ϵ were determined by psychrometric methods (for ref. see 3, 23) or by the pressure bomb technique (1, 6, 10, 17). L_p values have been obtained from swelling or shrinking experiments or by measuring the exchange of labeled water under certain experimental conditions and assumptions (*e.g.* at zero P as in plasmolytic experiments [11] or by assuming independent water exchange of tissue cells with extracellular space).

In the literature, L_p values for higher plant cells (3) differ by several orders of magnitude, probably because of different methods and models used for determination of L_p and its related parameters (18, 19, 23). The main objection to the models and methods used is that the quantities measured are mostly average values for bulk tissue. What is really required are measurements of P, ϵ, and L_p in single cells of higher plants. Up to now, these parameters have not been determined directly because of experimental difficulties. An important step in this direction was the development of the pressure probe 9 years ago (20), which has been applied to giant algal cells (13, 14, 21, 22, 24) and also to large cells of a higher plant (12, 16). This probe works on a compensation principle. Turgor pressure is transmitted via an oil-filled microcapillary, introduced into the cell, to a pressure transducer which transforms P into a proportional voltage. However, application of the probe has so far been limited by the size of the cells relative to the size of the apparatus, and by the compressibility of the silicon oil inside the apparatus. This problem was experienced by Meidner and Edwards (7), who essentially used the same measuring principle as Zimmermann and Steudle to determine P in stomatal guard cells. Meidner and Edwards were successful in applying pressure to guard cells (and to make them open or close), but not in recording pressure, probably because of the compressibility of their measuring device. In this paper, we present a miniaturized version of the pressure probe, which works on a compensation principle but has a very small, effective, compressible volume. With this device it is possible for the first time to measure directly and continuously pressure, cell volume changes (and, therefore, cell wall elasticity), and water flow in single tissue cells with diameters of 50 μm or less.

MATERIALS AND METHODS

Pressure Probe. The pressure probe (Fig. 1) consists of a microcapillary which is introduced into the cell, a pressure chamber containing the pressure transducer, and a motor-driven metal rod which can be used to change the volume of the pressure chamber. The whole device is filled with silicon oil. When the microcapillary is introduced into the cell, turgor pressure is transmitted by the silicon oil to the pressure transducer. However, turgor pressure compresses the oil or other compressible parts of the chamber (*e.g.* rubber of seals) to some extent, and thus the P recorded by the transducer will be reduced or even zero after puncturing the cell. This effect can be ignored if volume changes due to compression are negligible compared to changes in cell volume, *i.e.* if the volume of the apparatus is small.

For this reason, the effective volume of the pressure chamber must be drastically reduced if P and volume elasticity are to be measured in small plant cells (cell volume, $V_c = 10^{-2} - 10$ nl). This is achieved by fixing the oil/cell sap boundary (Fig. 1) in the very tip of the microcapillary so that the compressible volume of the whole device is that of the cell sap in the tip only (about 10^{-3}–10^{-1} nl). The boundary is adjusted and regulated by an electrical feedback mechanism. A silver wire (diameter, 1 μm at the end) is introduced into the tip of the microcapillary

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[2] Abbreviations: P: turgor pressure; ϵ: volumetric elastic modulus; L_p: hydraulic conductivity (water permeability).

as an internal electrode and another one is placed into the bathing solution.

A voltage of 20 to 50 mV is applied to the electrodes to measure the resistance. The resistance will depend on the position of the sap/oil boundary relative to the tip of the wire and will increase significantly when the wire becomes covered with oil. The boundary is adjusted by an automatically driven motor (Dunker motor GR 32.0 with a response time of 100 msec) to a reference resistance. The absolute value of the resistance depends on the tip diameter of both the microcapillary and the silver wire. The resistance varies between 50 and 200 MΩ, when the boundary is fixed at the capillary tip, and ranges from 0.5 to 20 MΩ, when it is located 100 μm behind the tip of the wire (Fig. 2). The resistance (2–200 MΩ) is measured at discrete time intervals (2–10 measurements per sec) or continuously with a frequency of 400 Hz. Changes in resistance are converted into controlling signals, which drive the motor. By this adjustment, not only the compressibility of the apparatus, but also the thermal expansion of the silicon oil (1.34×10^{-3} per degree) due to fluctuations in temperature can be eliminated. An extension of the silver wire due to thermal fluctuations has not been observed. The coefficient of linear thermal expansion of silver is about 10^{-6} per degree and, therefore, this effect should be negligible.

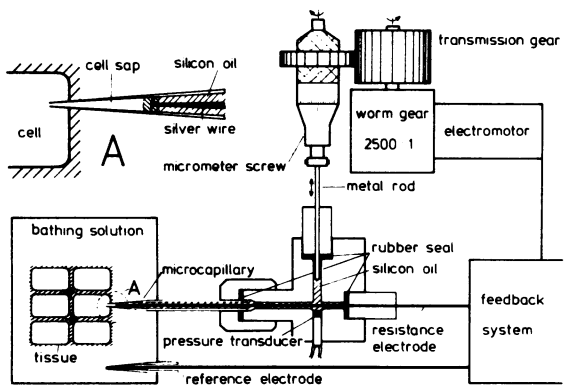

FIG. 1. Apparatus for measurement of hydrostatic pressure (P), hydraulic conductivity (L_p), and volumetric elastic modulus (ϵ) in tissue cells of higher plants. For further explanation see text.

Measurements of ϵ in plant cells by this compensation method also require that compression of oil in the apparatus be small compared to the volume changes of the cell. Volume compression of cell sap can be neglected because the compressibility coefficient of water is of the order of $10^{-4}\%$ bar^{-1}; this means that cell volume would change only $10^{-2}\%$/bar which is negligible. The effect of the compressibility of the apparatus on the determination of ϵ can be demonstrated in the following way. If the volume (and pressure) of the whole system is changed by moving the metal rod (Fig. 1), the volume change produced, dV_r (which can be calculated in the older version of the pressure probe [13, 21] from the feed of the rod and its diameter) will be equal to the sum of both the amount of compression of the oil, dV_{app}, and the expansion of the cell, dV_c:

$$\frac{dV_r}{dP} = \frac{dV_{app}}{dP} + \frac{dV_c}{dP} \quad (1)$$

The negative sign in equation 1 expresses that (due to compression) V_{app} decreases with increasing pressure, whereas V_r and V_c increase. Introducing the compressibility of the apparatus $\kappa_{app} = -\frac{1}{V_{app}}\frac{dV_{app}}{dP}$ and expressing $\frac{dV_c}{dP}$ by the elastic modulus of the cell $\epsilon = V_c \frac{dP}{dV_c}$ equation 1 yields:

$$\frac{1}{V_c}\frac{dV_r}{dP} = \frac{V_{app}}{V_c}\kappa_{app} + \frac{1}{\epsilon} \quad (2)$$

Equation 2 gives the limits for the determination of ϵ and the conditions for constructing the pressure probe. It states that if $\epsilon \ll \frac{V_c}{V_{app}}\frac{1}{\kappa_{app}}$ a good approximation of ϵ will be given by $V_c \frac{dP}{dV_r}$. On the other hand, if the ratio V_{app}/V becomes very large (small cells) the first term on the right side of equation 2 will be of the same order as $\frac{1}{V_c}\frac{dV_r}{dP}$. In this case the error in the determination of ϵ according to equation 2 will be large.

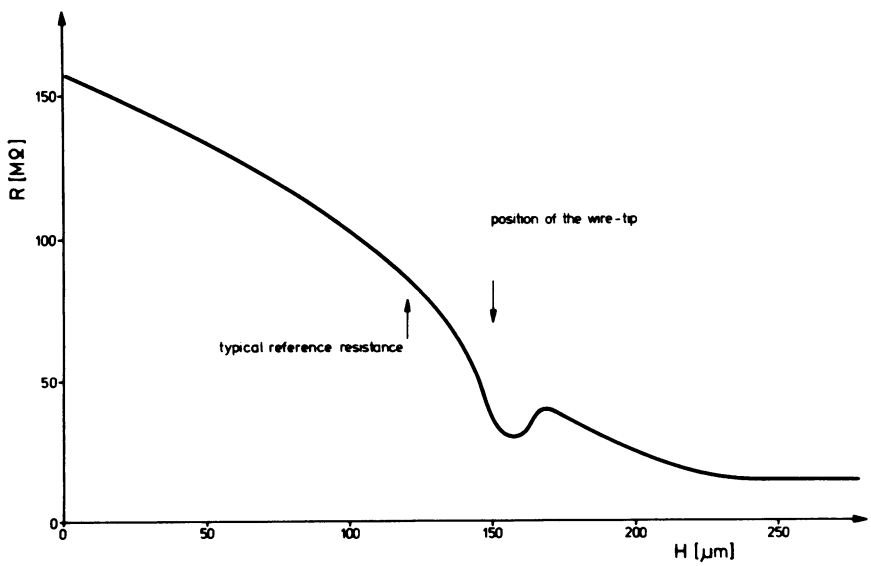

FIG. 2. Electrode resistance of the pressure probe shown in Figure 1 as a function of the distance of the oil/cell sap boundary from capillary tip (H). For resistance characteristic shown in the figure the capillary tip was 1 μm in diameter. Tip of silver wire was about 2 μm in diameter and located about 150 μm behind capillary tip. Because of differences in surface tension and of different adhesion of silicon oil and water to electrode surface, relatively large changes in resistance occurred when the boundary moved across the wire tip. A typical reference resistance to fix the boundary would correspond to a position about 20 to 30 μm in front of the tip of the wire.

For the apparatus shown in Figure 1, V_{app} (*i.e.* the tip volume) is 10^{-3} – 10^{-1} nl and κ_{app} is approximately that of water (about 10^{-4} bar^{-1}). If V_c is 10^{-2} to 10 nl (cell diameter of 25-250 μm) and ϵ 10 to 100 bar (3), then $\frac{V_{app}}{V_c}\epsilon_{app}$ will be 10^{-5} bar^{-1} or less and thus negligible as compared to $1/\epsilon$ (0.1-0.01 bar^{-1}).

Experimental Procedure. Prior to measurement, the boundary silicon oil/water was adjusted at the tip of the wire or at a certain point (*i.e.* a certain resistance was fixed). Tissue slices from pepper fruits (*Capsicum annuum*) about 20 mm in length and 2 mm in height were incubated for several hr in Johnson solution modified after Epstein (4) with the following composition: 3 mM KNO_3, 2 mM $Ca(NO_3)_2$, 1 mM $NH_4H_2PO_4$, and 0.5 mM $MgSO_4$. They were then transferred into a small chamber containing well stirred Johnson solution at a constant temperature (20 C). Tissue slices were fixed on a bed of very small nails to prevent any movement of the slices due to volume changes in response to turgor regulation processes during measurement. The pressure probe was introduced with a Leitz manipulator under a stereomicroscope (magnification: 40 – 280). Ten to 20 min after puncturing the cell, a constant P of 2 to 4 bar could be recorded over a period of 5 hr and more. To determine ϵ, the oil/cell sap boundary in the tip of the microcapillary was moved by certain amounts, and the instantaneous changes in cell turgor pressure, ΔP, were recorded (see Fig. 4). Changes in the cell volume, ΔV_c, were calculated by taking these regions of the capillary tip as truncated cones. The movement of the boundary was measured under the microscope. To evaluate the (inner) diameters of the microcapillary at different distances from the tip the dimensions of a series of the commerical microcapillaries used (Transidyne General, Ann Arbor, Mich.) were determined under a microscope with high magnification (600×). A relation was derived between the inner diameter (D) and the distance (H) from the capillary tip, which is given by:

$$D(H) = 0.8\,\frac{D_0 - d_0}{L_0}\,H + 0.2\left(d_0 + \frac{(D_0 - d_0)\,\tan^{-1}(H - L_0/3)}{\tan^{-1}(2L_0/3) + \tan^{-1}(L_0/3)}\right) \quad (3)$$

where D_0 = inner diameter of glass tubes used for preparing micropipettes; d_0 = inner diameter of microcapillary tip; L_0 = length of capillary tip. It is shown in Figure 3 that the calculated values of the inner diameter (D) as a function of the distance from the capillary tip (H) are in good agreement with the measured values.

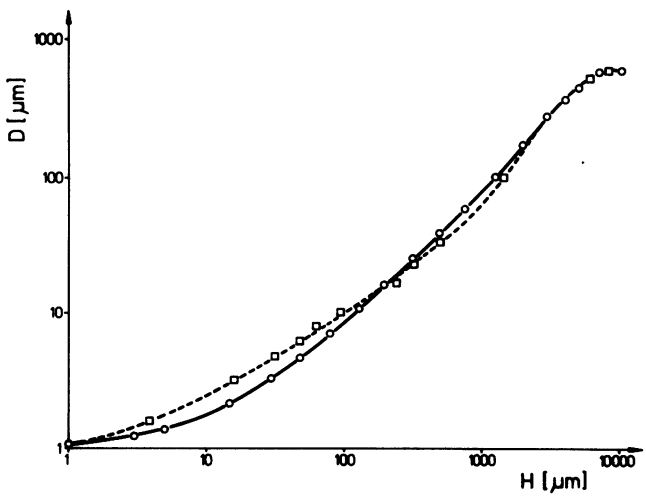

FIG. 3. Inner diameter (D) of microcapillaries used as pressure probes as a function of distance (H) from microcapillary tip. (○): D values calculated from equation 3 (D_0 = 600 μm; d_0 = 1 μm; L_0 = 7,000 μm); (□): average values on 10 micropipettes measured under the microscope at a magnification of 600×.

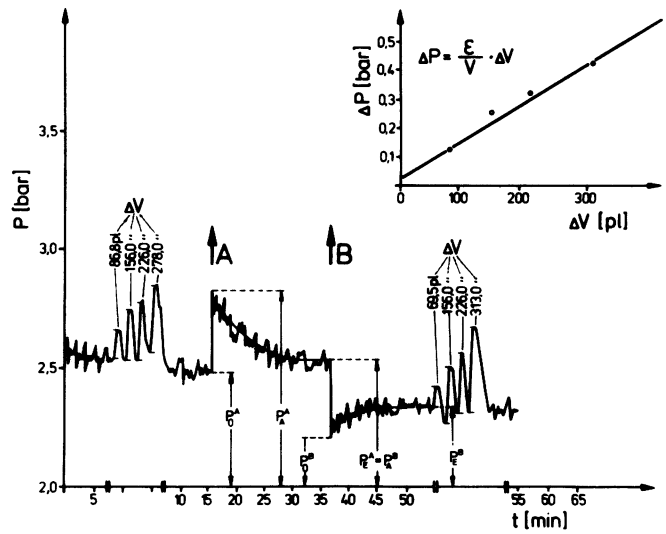

FIG. 4. Time course of change in cell turgor pressure (P) of a fruit tissue cell of *Capsicum annuum* after sudden increase (A) and decrease (B) of stationary turgor pressure. P changes exponentially with time until a new level of stationary P is reached. ϵ is determined from the dependence of initial changes in turgor pressure (ΔP) upon rapid cell volume changes (ΔV) that occur prior to volume flow. In the inset, ΔP is plotted *versus* ΔV for a cell with a volume of V = 11 nl. ϵ, can be calculated from slope of straight line, using equation 4.

The movement of the boundary was performed in about 1 sec or less, *i.e.* in a time which was short compared to the half-time of water exchange (Fig. 4). Tests with the tip in the bathing medium ensured that errors due to shockwave pressures during the pressure changes could be neglected for the tip diameters used (1-3 μm).

Capillary forces were small, because the oil/cell sap boundary was fixed at a position where the diameter of the capillary was relatively large (5-50 μm). By using the electronic feedback mechanism the boundary oil/cell sap could be fixed within ±5 to 10 μm, which means that the cell volume could be kept constant within about 10^{-7} to 10^{-5} μl (inner diameter of the capillary at the boundary 5-50 μm).

Measurement of cell dimensions under the microscope, taking the shape of the cell to be approximately ellipsoid or cuboid, allowed determination of the cell volume, V_c. The error in these determinations was of the order of 15 to 20%. Measurement of ϵ of cells of *Valonia utricularis* with the modified pressure probe yielded exactly the same results as with the older version of the pressure probe introduced by Zimmerman *et al.* (13, 20) and normally used for measurements in *Valonia* cells (13, 21, 24).

RESULTS AND DISCUSSION

Figure 4 shows recordings of turgor pressure and ϵ measurements in a cell located in the first cell layer of a tissue slice of *C. annuum* (100-300 μm in size). As soon as P has reached a constant value after introduction of the microcapillary, the cell volume is changed by a defined amount, ΔV (0.5-5% of the cell volume which ranged from 2 to 12 nl) by shifting the oil/cell sap boundary in the tip of the capillary (Fig. 4). ϵ can be calculated from the corresponding changes in cell turgor, using the equation of Philip (9):

$$\epsilon = V \cdot \frac{dP}{dV} \simeq V \cdot \frac{\Delta P}{\Delta V} \quad (4)$$

For estimation of water flow, the stationary turgor pressure is changed by shifting the oil/cell sap boundary to a new reference point. The magnitude of water flow induced by the change in cell P can be calculated from the turgor pressure relaxation if ϵ is known (13, 14, 21, 22).

The ϵ values obtained (Fig. 5, a and b) were of the order of 2 to 25 bar and strongly dependent on P which could be lowered or increased by varying the external osmotic concentration with NaCl or sucrose. Figure 5, a and b also demonstrates that ϵ is a function of cell volume (size). Similar effects of pressure and volume have been observed for giant algal cells (21, 22, 24) and also for a higher plant cell (16) by using the unmodified version of the pressure probe, although the absolute value of ϵ was larger by an order of magnitude for the algal cells. The results suggest that the averaged values of ϵ obtained by psychrometry and by the pressure bomb technique should be treated with caution. Water transport in higher plant tissues, which is determined by wall elasticity, is a rather complex phenomenon controlled by both P and cell volume (size).

It could be argued that the volume dependence of ϵ results partly from a region of high elastic extensibility created around the tip of the pressure probe by puncturing the cell. We do not feel that this objection is serious as this region would only constitute about $^1/_{500}$ of the total geometric surface of a tissue cell. The ϵ value of this region would have to be extremely low to bring about a change in the over-all ϵ value, and indeed, large extension of the wall in the tip region should be observable under the microscope, which is not the case. Furthermore, both versions of the pressure probe were used for measurement on giant algal cells with a wide range of tip diameters (5–100 μm), but a dependence of ϵ on the tip diameter was never observed.

The pressure probe provides a new tool for measuring not only pressure and wall elasticity, but also the rate of water exchange between individual cells and the surrounding tissue. Figure 6a reveals that water flow, J_V, across the membrane of a tissue cell in response to changes in P (at point A and B in Fig. 4) is time-dependent.

As shown in Figure 6b, where log J_V is plotted against time, water flow (as turgor pressure) decreases exponentially with a half-time $T_{1/2}$, of about 4 min. Assuming that the cell is exchanging freely with the external medium, *i.e.* that it behaves like a single cell in a large medium, this value can be used for a first (very rough) approximation of L_p. In this case, L_p would assume a value of $5 \cdot 10^{-7}$ cm sec^{-1} bar^{-1}. However, the calcula-

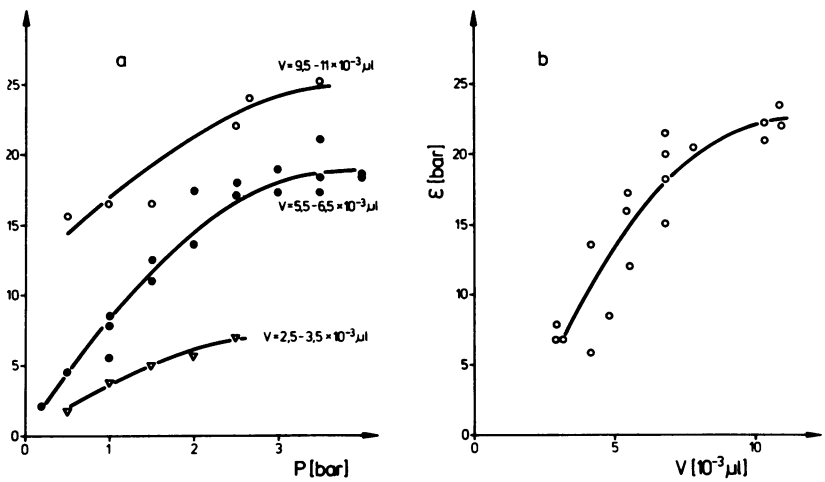

FIG. 5. a: Effect of cell turgor pressure (P) on the volumetric elastic modulus (ϵ) of fruit tissue cells of *Capsicum annuum*. Pressure dependence of ϵ is shown for three different intervals of cell volume (V). b: Volume dependence of ϵ for the same cells as in Figure 5a shown for pressure interval between 2.25 and 2.75 bar.

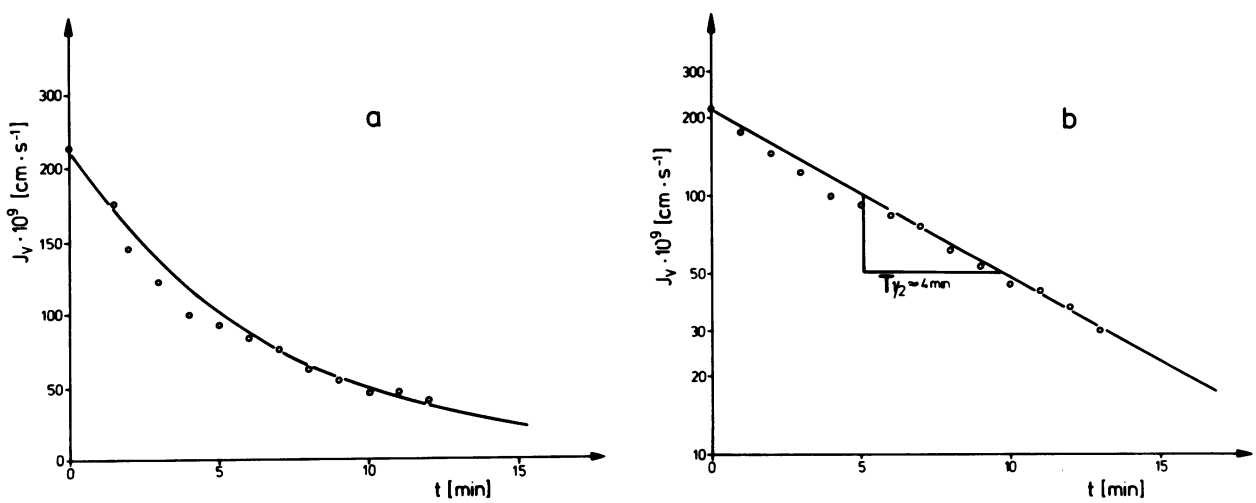

FIG. 6. Time dependence of the volume flow [$J_v(t)$] in a single tissue cell of *Capsicum annuum* fruit after artificial change of turgor pressure (ΔP) of about 0.45 bar at t = 0 (Fig. 6a). $J_v(t)$ was calculated from curve A in Figure 4. Semilog plot of $J_v(t)$ against time (Fig. 6b) yields a straight line. From slope of this line, the half-time for water exchange can be calculated to be $T_{1/2}$ = 4 min.

tion of L_p is not straightforward for a cell within a tissue, since the total hydraulic resistance to water flow will also include the resistance of the apoplasmic barrier and the resistance of the surrounding cells (for further discussion see refs. 3, 8, 23). More data will have to be collected to estimate contributions of these different pathways and to achieve an exact evaluation of L_p of cell membranes. This could probably be done by measuring the propagation of P in a tissue with two or more probes located in different cells, or by determining the dependence of water flux equilibration as a function of depth of a cell in a tissue slice.

The advantages of the method presented here are that ϵ and P can be determined directly in single cells without any further assumptions. P can be recorded continuously over long periods of time (5 hr and more) with a high accuracy ($3\text{–}5 \cdot 10^{-2}$ bar). Errors due to leakages around the tip during the experiment can be ruled out, since the equipment is very sensitive to leakages (which for example, may be caused by vibrations) which produce sudden drops in the pressure.

A definite limitation for application of the method to cells in plant tissues is given by the ratio of the hydraulic resistance of the cell membrane ($1/L_p \cdot A$) to the resistance of the capillary tip. When the resistance of the tip to water flow or the time constant of water exchange across the tip approaches the same order of magnitude as the corresponding membrane parameters, the time course of P relaxation processes in response to a disturbance of the osmotic equilibrium can no longer be determined accurately. In the experiments presented here, the time constant of water exchange across the tip was about 1 sec (tip diameter $\simeq 1$ μm), and thus two orders of magnitude smaller than the half-time of the water exchange across the cell membrane (4 min). A further reduction of the tip diameter which might be necessary for measurements in very small cells (diameter < 20 μm) is not only prohibitive because of the increase in the tip resistance, but also because of danger of shockwave pressure and the blocking of the tip by cytoplasmic material.

The equipment excludes possible sources of error which may arise when ϵ values are determined by the pressure bomb technique (1). These errors include uncertainty in determination of the plasmolytic volume, differences in the osmotic concentration, volume, and ϵ values of the cells, and the inaccuracy in the estimation of volume and pressure changes in the nonlinear region of the "pressure-volume" curve.

Very recently Ferrier and Dainty (5) published a new method for determining ϵ (and L_p) in higher plant cells. The method is based on an elastic deformation of a cell layer by an external force. Assuming that at small deformations, the elastic expansion of the cell in each of the three dimensions is independent and that all cells in the layer behave elastically the same, the authors obtained a linear relationship between cell thickness and applied (uniaxial) force, from which ϵ could be estimated to be of the order of 2 bar for fully turgid onion epidermal cells. With further assumptions (*e.g.* free exchange of water between cell and medium, which is a rather rough assumption under the given experimental conditions) they arrive at L_p values of 2.3 to $5.1 \cdot 10^{-6}$ cm sec^{-1} bar^{-1}. If their assumptions are correct, their method would be a new approach, but would apparently yield averaged values of ϵ for thin tissue slices. Our method is suited for measurements on single cells, even if they are deep inside a tissue.

We would also like to mention another method for direct measurement of P in small plant cells, which seems to be promising at first sight (Zimmermann, unpublished data). This technique is based on the introduction of a small gas bubble into cells of a tissue with subsequent determination of pressure changes within the cells from the corresponding volume changes of the gas bubble (Boyle-Mariotte's law). Gas bubbles can be introduced into cells by means of very fine capillaries or electrode processes. Preliminary experiments on *V. utricularis* have shown that this method works in principle, but that a greater technical effort is required to measure pressure changes with the same accuracy as the pressure probe introduced here. The simplest version of the gas bubble pressure probe is not sufficiently sensitive for the measurement of pressure changes of the desired order of magnitude. In a first approximation, the pressure in the gas bubble is inversely proportional to the third power of the radius, so that in the physiological pressure range the changes in diameter to be measured under the light microscope are very small (0.1 μm) in relation to the resolution of the microscope (about 0.5 μm). The sensitivity may be improved, if resolution is increased by using advanced optical techniques. Furthermore, with this particular method of measuring pressure changes, solubility of the gas in the cell sap and diffusion of gas through the membrane barrier have to be taken into account, because in time both processes lead to a change in the volume of gas inside the cell. In principle, it would be possible to overcome these problems by using sparingly soluble gases. This technique, which could represent an elegant way for measuring pressure changes in a number of cells simultaneously, would most certainly require more technical developments in order to be of practical use.

We feel that the pressure probe presented here is at the present the simplest way of measuring pressure in tissue cells of higher plants with a minimum number of assumptions. Possibly, the pressure probe will not only be useful for the study of swelling and shrinking processes and water transport in higher plant tissues, but also for investigation of both the regulation of stomatal responses (by direct measurement of guard cell pressure), and of pressure-regulated membrane transport in higher plants, which has been shown to exist in giant algal cells (2, 15, 21, 24).

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LITERATURE CITED

1. Cheung YNS, MT Tyree, J. Dainty 1976 Some possible sources of error in determining bulk elastic moduli and other parameters from pressure-volume curves of shoots and leaves. Can J Bot 54: 758–765
2. Coster HGL, E Steudle, U Zimmermann 1976 Turgor pressure sensing in plant cell membranes. Plant Physiol 58: 636–643
3. Dainty J 1976 Water relations of plant cells. In U Lüttge, MG Pitman, eds, Encyclopedia of Plant Physiology, New series, Vol 2, Part A. Springer-Verlag, Berlin, pp 12–35
4. Epstein E 1972 Mineral Nutrition of Plants: Principles and Perspectives. John Wiley & Sons, New York
5. Ferrier JM, J Dainty 1977 A new method for measurement of hydraulic conductivity and elastic coefficients in higher plant cells using an external force. Can J Bot 55: 858–866
6. Hellkvist J, GP Richards, PG Jarvis 1974 Vertical gradients of water potential and tissue water relations in Sitka spruce trees measured with the pressure chamber. J Appl Ecol 11: 637–667
7. Meidner H, M Edwards 1975 Direct measurement of turgor potential of guard cells. I. J Exp Bot 26: 319–330
8. Molz FJ, E Ikenberry 1974 Water transport through plant cells and cell walls: theoretical development. Soil Sci Soc Am Proc 38: 699–704
9. Philip JR 1958 The osmotic cell, solute diffusibility, and the plant water economy. Plant Physiol 33: 264–271
10. Scholander PF, HT Hammel, EA Hemmingsen, ED Bradstreet 1964 Hydrostatic pressure and osmotic potential in leaves of mangroves and some other plants. Proc Nat Acad Sci USA 52: 119–125
11. Stadelmann E 1966 Evaluation of turgidity, plasmolysis and deplasmolysis of plant cells. *In* DM Prescott, ed, Methods in Plant Physiology, Vol II. Academic Press, New York, pp 143–216
12. Steudle E, U Lüttge, U Zimmermann 1975 Water relations of epidermal bladder cells of the halophytic species *Mesembryanthemum crystallinum*: direct measurement of the hydrostatic pressure and hydraulic conductivity. Planta 126: 229–246
13. Steudle E, U Zimmermann 1971 Hydraulische Leitfähigkeit von *Valonia utricularis*. Z Naturforsch 26b: 1302–1311
14. Steudle E, U Zimmermann 1974 Determination of the hydraulic conductivity and of reflection coefficients in *Nitella flexilis* by means of direct cell-turgor pressure measurements. Biochim Biophys Acta 332: 399–412
15. Steudle E, U Zimmermann, PI Lelkes 1977 Volume and pressure effects on the potassium fluxes of *Valonia utricularis*. *In* M Thellier, ed, Transmembrane Ionic Exchange in Plants. CNRS, Paris. In press
16. Steudle E, U Zimmermann, U Lüttge 1977 Effect of turgor pressure and cell size on

the wall elasticity of plant cells. Plant Physiol 59: 285–289
17. Tyree MT, HT Hammel 1972 The measurement of turgor pressure and water relations of plants by pressure bomb technique. J Exp Biol 24: 267–282
18. Zimmermann U 1977 Cell turgor regulation and pressure mediated transport processes. *In* D. Jennings, ed, Proceedings of 3rd Symposium of the Society of Experimental Biology. Integration of Activity in Higher Plant. Cambridge University Press. In press
19. Zimmermann U 1978 Physics of turgor regulation in plants. Annu Rev Plant Physiol. In press
20. Zimmermann U, H Räde, E. Steudle 1969 Kontinuierliche Druckmessung in Pflanzenzellen. Naturwissenschaft 56: 634.
21. Zimmermann U, E Steudle 1974 The pressure-dependence of the hydraulic conductivity, the membrane resistance and membrane potential during turgor pressure regulation in *Valonia utricularis*. J Membr Biol 16: 331–352
22. Zimmerman U, E Steudle 1975 The hydraulic conductivity and volumetric elastic modulus of cells and isolated cell walls of *Nitella* and *Chara*: pressure and volume effects. Aust J Plant Physiol 2: 1–12
23. Zimmermann U, E Steudle 1978 Physical aspects of water transport. Adv Bot Res. In press
24. Zimmermann U, E Steudle, PI Lelkes 1976 Turgor pressure regulation in *Valonia utricularis*: effect of cell wall elasticity and auxin. Plant Physiol 58: 608–613